



UNIVERSITÀ DEGLI STUDI DI TORINO

**DIPARTIMENTO DI SCIENZE VETERINARIE**

**Settore di Clinica Medica Veterinaria**

**INHALED MINERAL PARTICLES IN  
EQUINE ASTHMA: INNOCENT  
BYSTANDER OR CONCURRENT CAUSE  
OF DISEASE?**

**Alessandra Romolo, Jean-Pierre Lavoie and Michela Bullone**



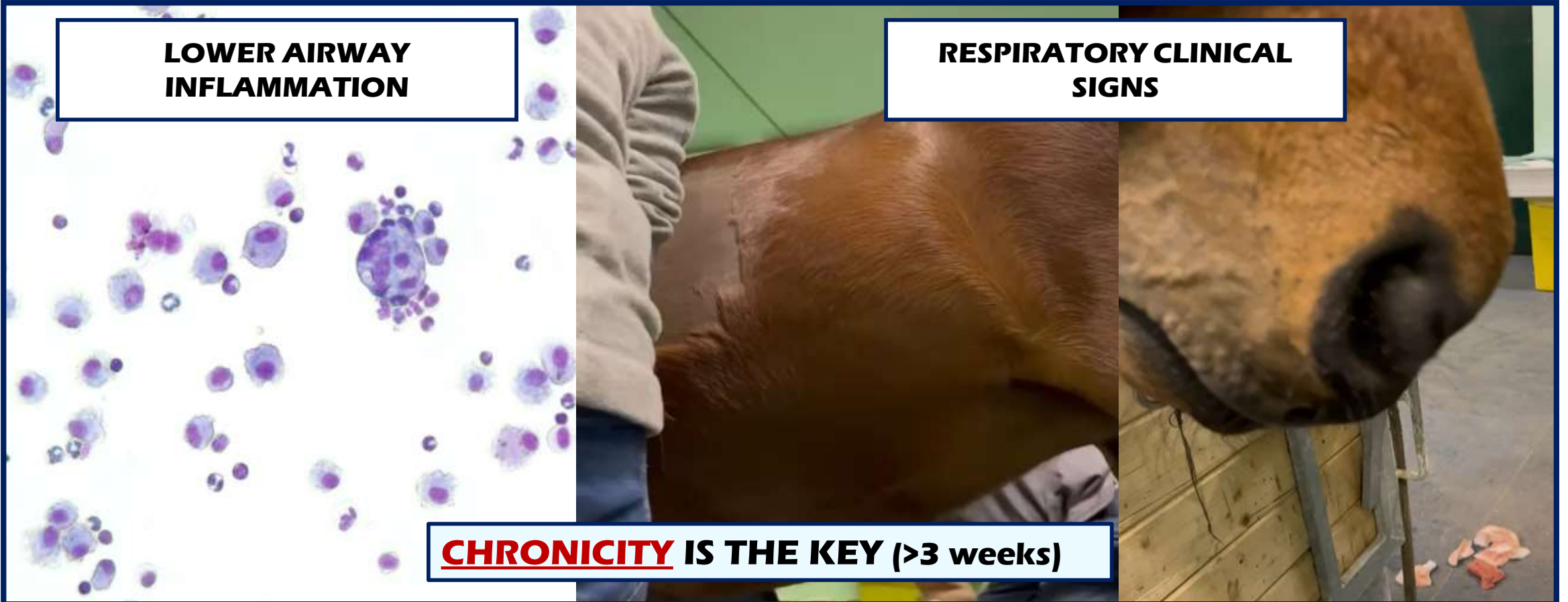
# EQUINE ASTHMA

**HIGHTLY PREVALENT DISEASE IN THE EQUINE ADULT POPULATION**

**LOWER AIRWAY  
INFLAMMATION**

**RESPIRATORY CLINICAL  
SIGNS**

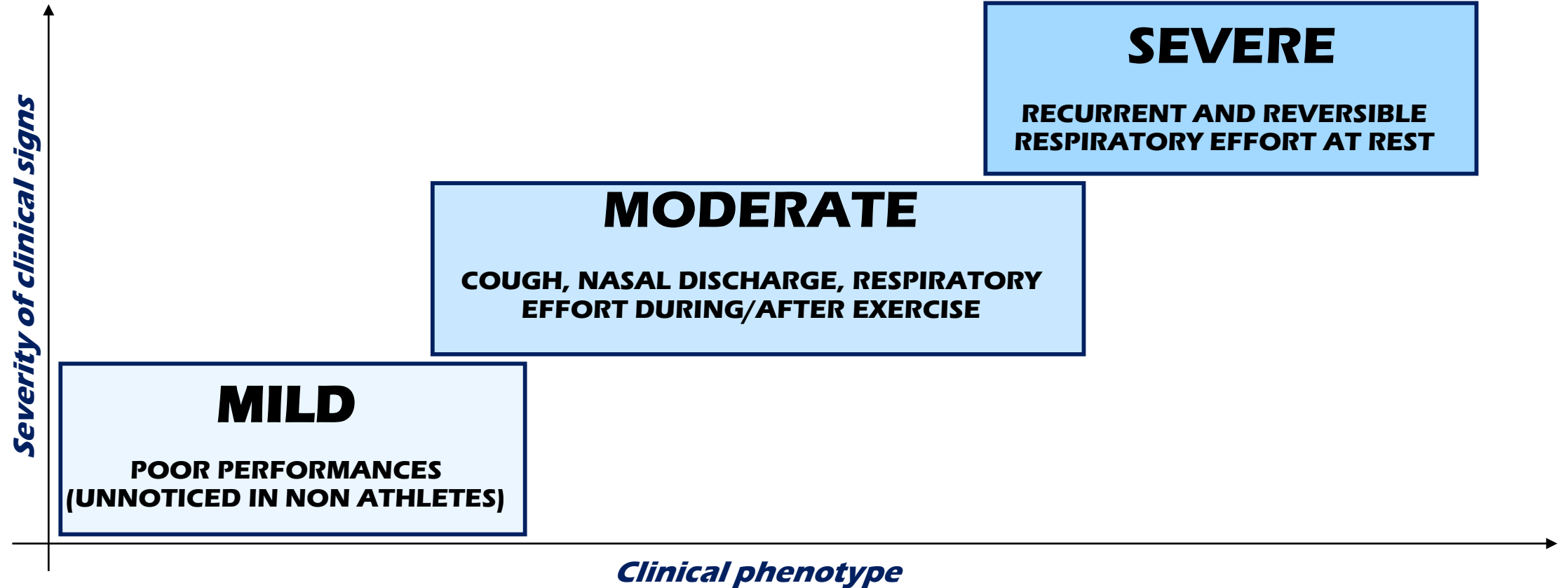
**CHRONICITY IS THE KEY (>3 weeks)**



# EQUINE ASTHMA



## DIFFERENT PHENOTYPES OF DISEASE

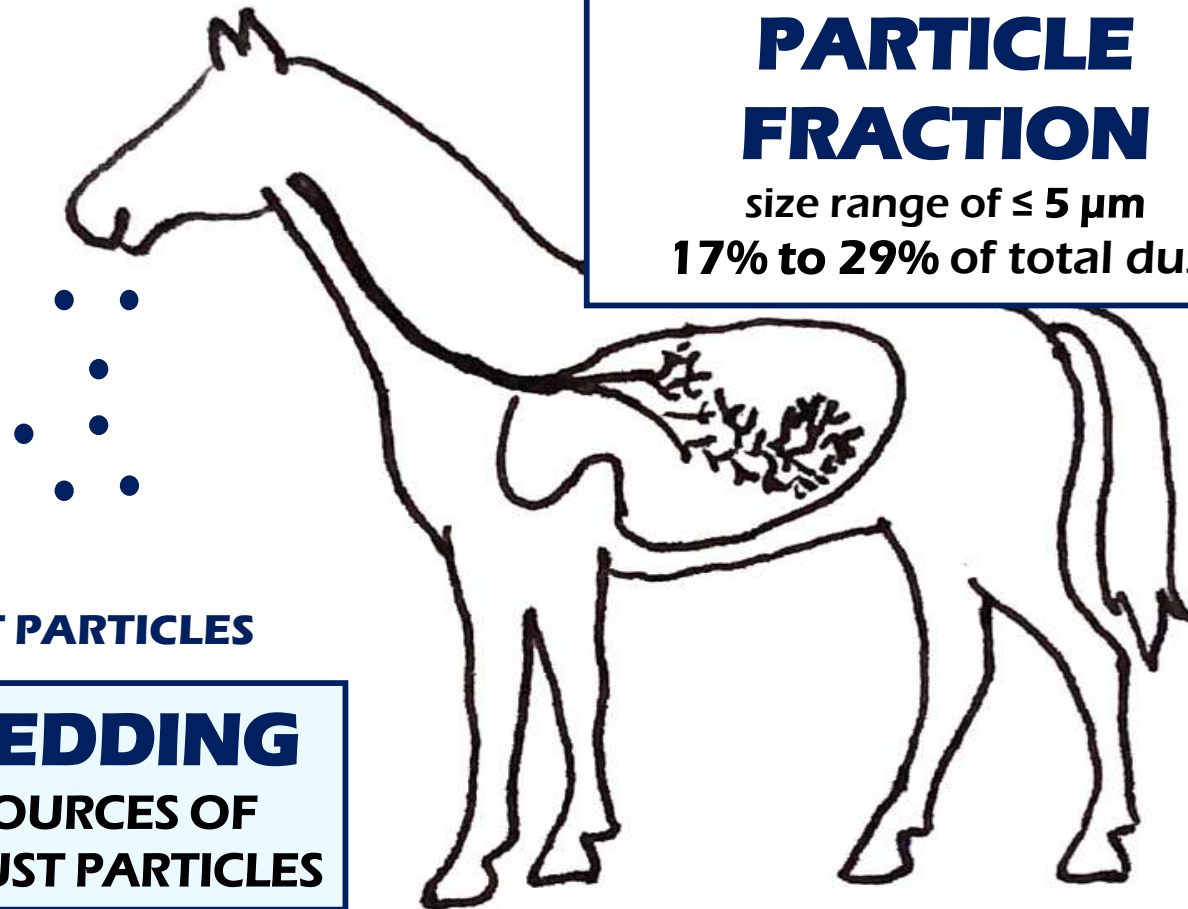


# PATHOGENESIS



## RESPIRABLE PARTICLE FRACTION

size range of  $\leq 5 \mu\text{m}$   
17% to 29% of total dust



DUST PARTICLES

## HAY & BEDDING

THE MAIN SOURCES OF  
RESPIRABLE DUST PARTICLES



# RATIONALE OF OUR WORK



**HORSES ARE EXPOSED TO  
UNRECOGNIZED HIGH  
LEVELS OF INHALED  
MINERAL PARTICLES**

**THIS CAN  
CONTRIBUTE TO  
RESPIRATORY  
PATHOLOGY**



# **RATIONALE FOR PERFORMING THE STUDY**

## **HUMANS**

**Pulmonary diseases caused by organic and inorganic dusts are a significant source of occupational morbidity and mortality**

**The epidemiological evidence reinforces the need for prompt recognition and control of the respirable agents**

**Instructors and trainers are exposed to this dust for many hours daily, especially if the horse riding is done in indoor arenas**



# RATIONALE FOR PERFORMING THE STUDY

## SILICA DUSTS

**SiO<sub>2</sub>**

**PATHOGENICITY**

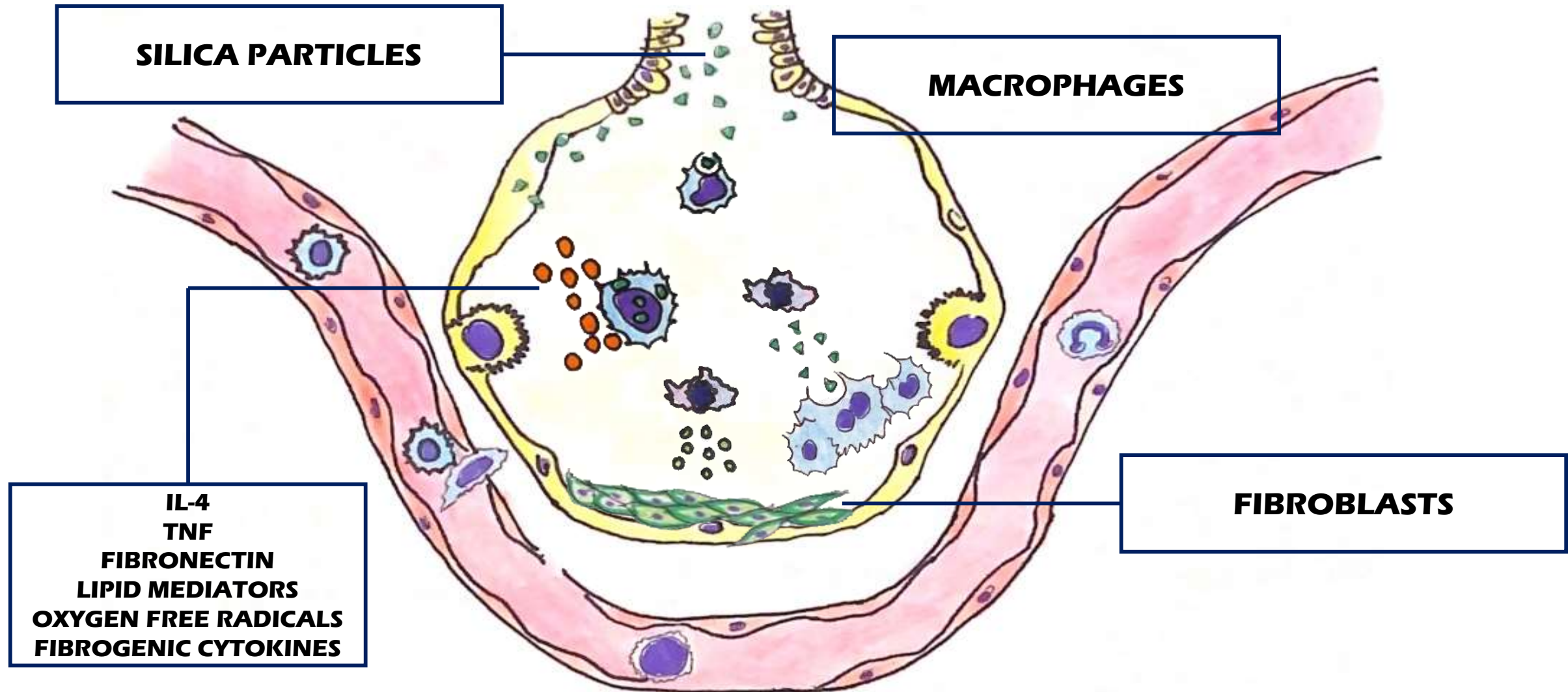
**crystalline silica**

**+++**

**amorphous silica**



# SILICA-INDUCED INFLAMMATORY PATTERN







# **HYPOTHESIS AND OBJECTIVES**

## **HYPOTHESIS**

**INHALED MINERAL DUSTS  
ARE INVOLVED IN EQUINE  
ASTHMA  
PATHOPHYSIOLOGY**

## **OBJECTIVES**

**TO QUANTIFY AND COMPARE  
PARTICLES OF CRYSTALLINE  
SILICON DIOXIDE IN  
RESPIRATORY SECRETIONS**



# MATERIALS AND METHODS

**42 cases**  
**13 SEA, 19 MEA, 10 controls**

**BAL PROCEDURE**

**CYTOSPIN SLIDES  
PREPARATION**

**CYTOSPIN SLIDES  
OBSERVATION**

**Veterinary Teaching Hospital of the  
University of Turin and Montréal**

**Cross sectional study performed on cases searched on:**

- ❖ **April 2021 and prospectively included starting from April 2021 ( Turin OVU )**
- ❖ **November 2021 (Montréal OVU)**

- **informations on history**
- **first examination**
- **cytology slides still available**

- **other respiratory diseases**
- **no medical history reported**
- **uncertain diagnosis**
- **TTW: positive bacterial culture**
- **corticosteroids/antimicrobial treatments in the 2-week period preceding BALF**



## **MINERAL PARTICLE COUNTS**

- ❖ **COUNTS PERFORMED ON 30 HPF PER SLIDES (40X), OPTICAL MICROSCOPY WITH POLARIZED LIGHT**
- ❖ **ALL THE SLIDE INSPECTED AND MINERAL PARTICLE COUNT EXPRESSED PER HIGH POWER FIELD (INTRA AND EXTRACELLULAR)**

**SILICA PARTICLES CLASSIFIED BASED ON**

**COLOUR (milky appearance)**

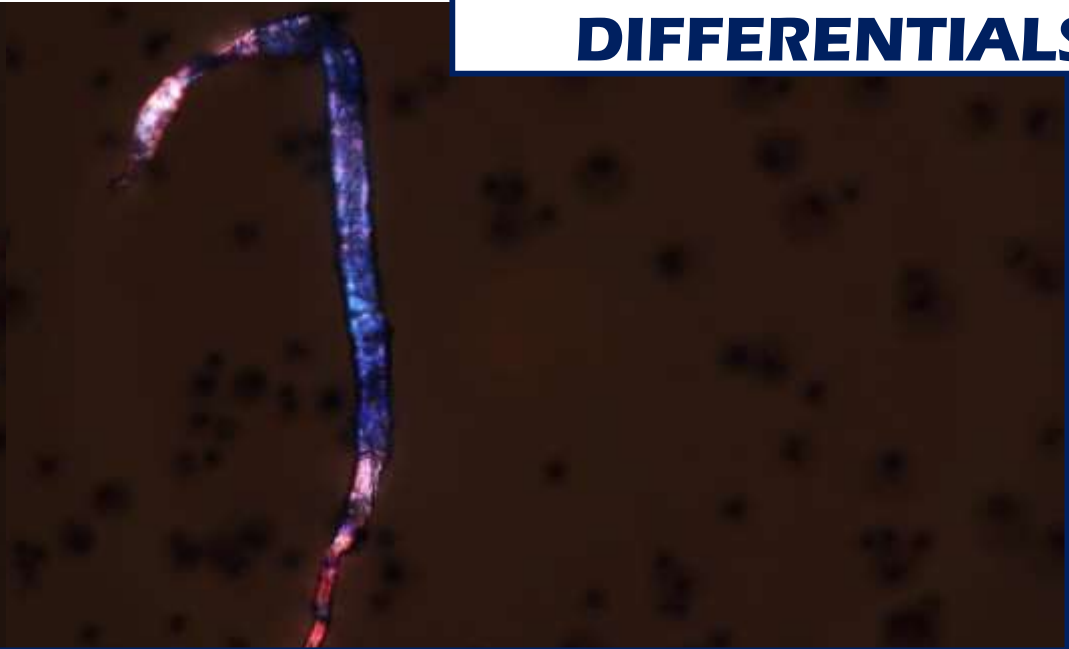
**SIZE  $\leq 2,5 \mu\text{m}$**

**LOCALIZATION**

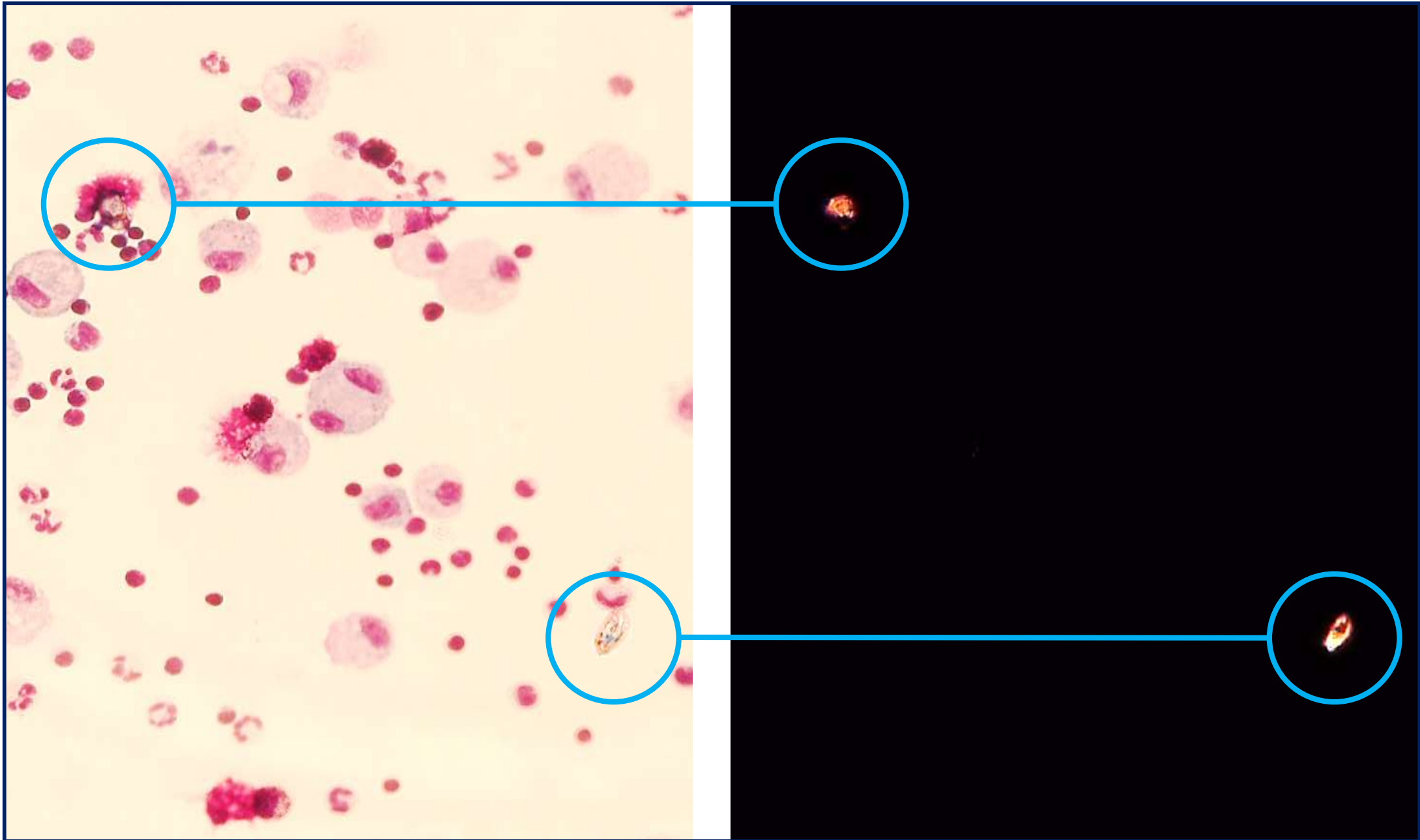
**Silicon dioxide within a macrophage in a horse with MEA (40x).**



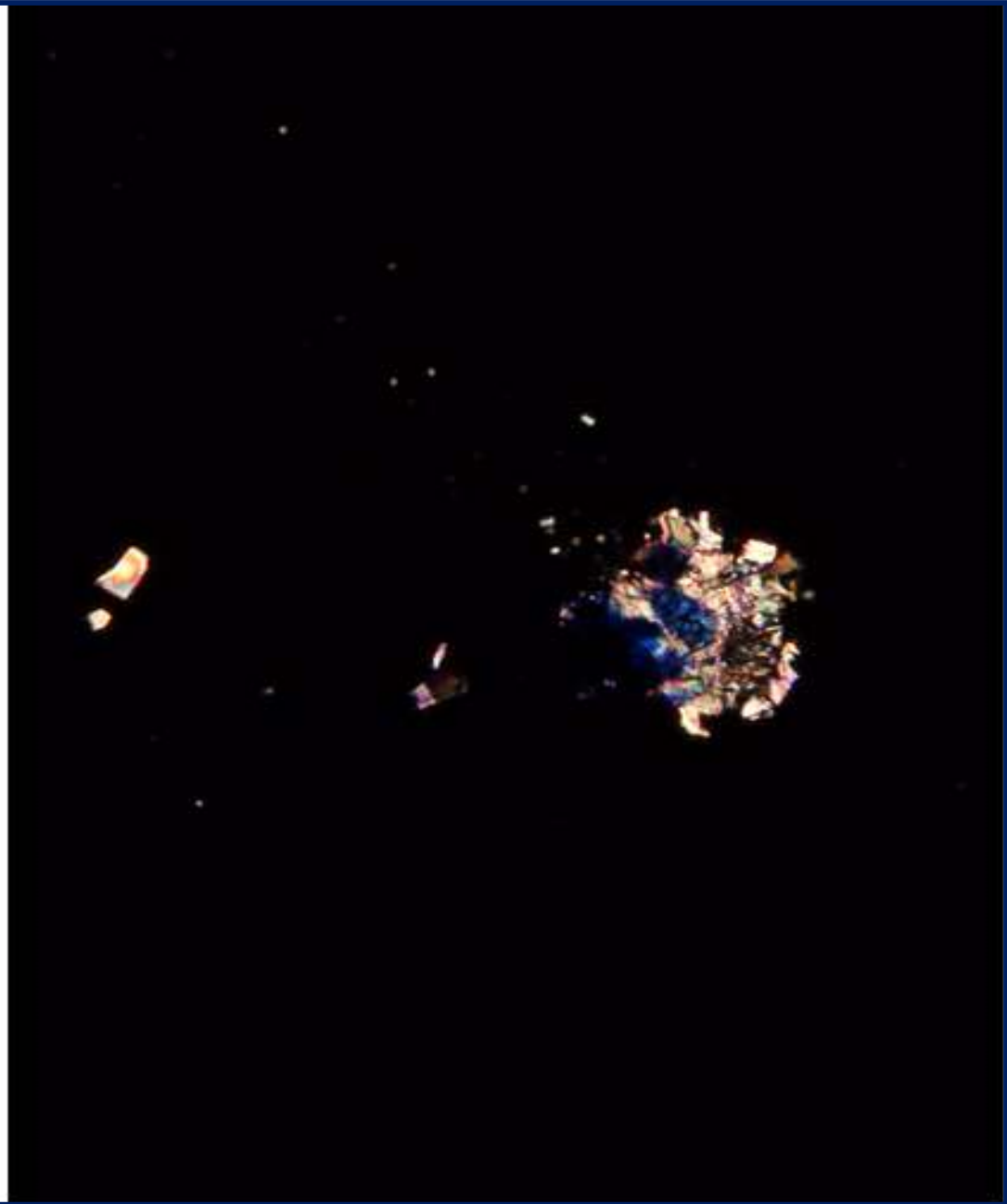
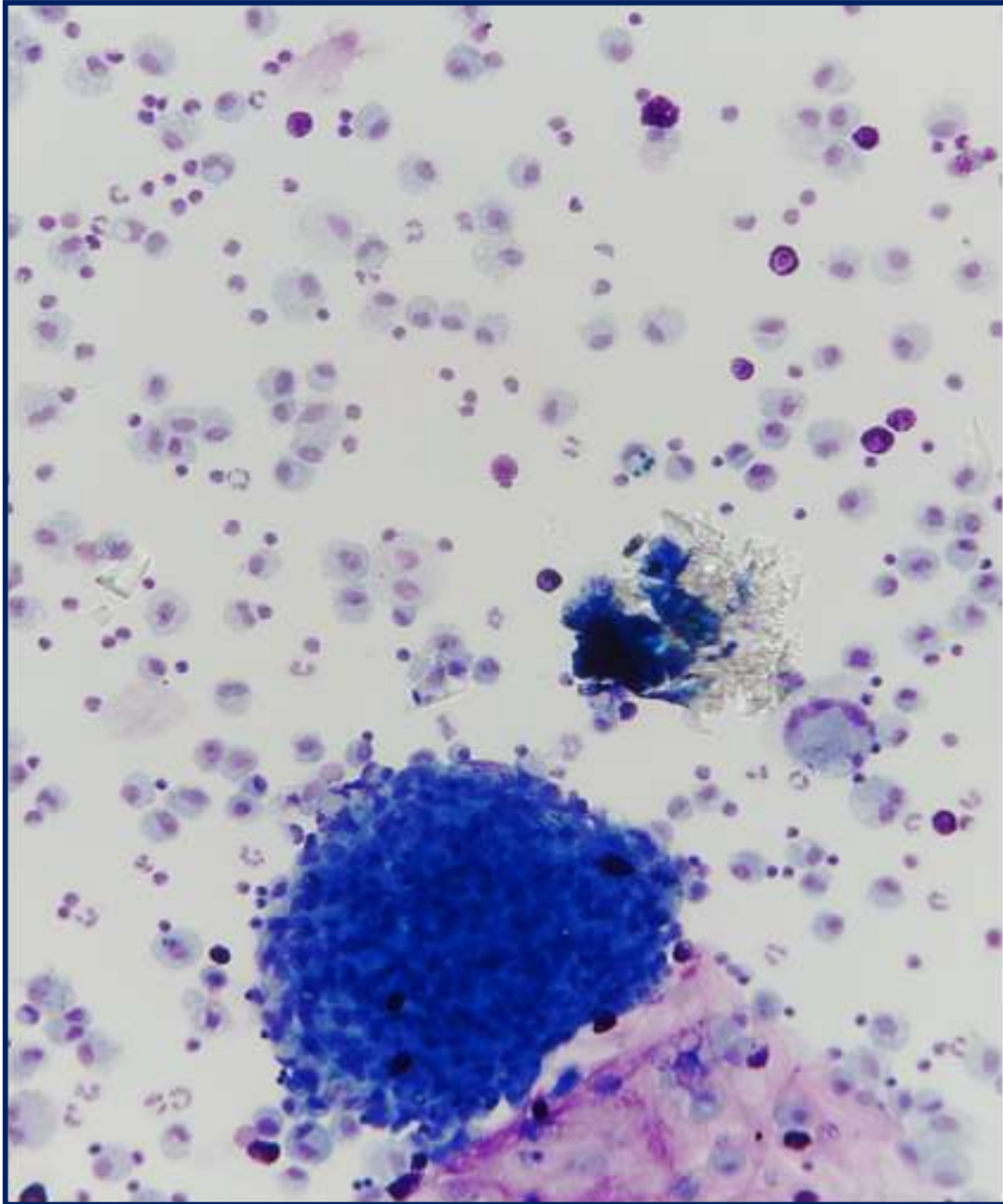
## SILICA PARTICLES DIFFERENTIALS



Vegetable elements, probably hay, in a horse with MEA (40x).



**Accumulation of large amounts of iron by lysosomes in a horse with MEA (40x).**

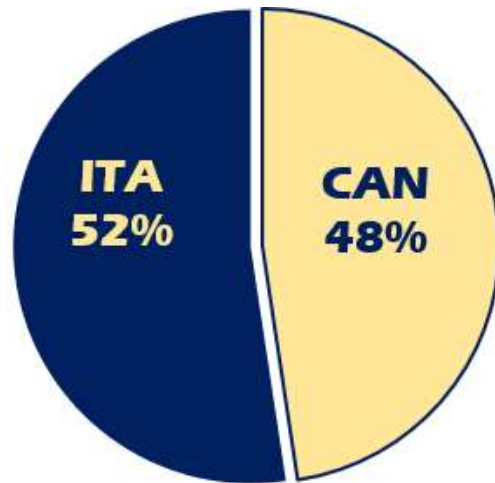


**Composite inhaled minerals probably characterized by silicon dioxide microparticles in a horse with MEA (40x).**



# RESULTS

## HORSES



	Turin cohort (N=22)	Montreal cohort (N=20)
Controls		
N	0	10
Sex (M:F)	-	<u>5</u> :5
Age [yrs]	-	10.4 ± 3.6
Mild to moderate equine asthma (MEA)		
N	14	5
Sex (M:F)	8:6	<u>3</u> :2
Age [yrs]	7.4 ± 5.7	8.2 ± 1.6
Severe equine asthma (SEA)		
N	8	5
Sex (M:F)	<u>6</u> :2	<u>3</u> :2
Age [yrs]	15.6 ± 5.4	16.0 ± 4.5

**BREED** → HANNOVER , QUARTER HORSE,  
STANDBRED

**BAL** → NOVEMBER – FEBRUARY - MARCH

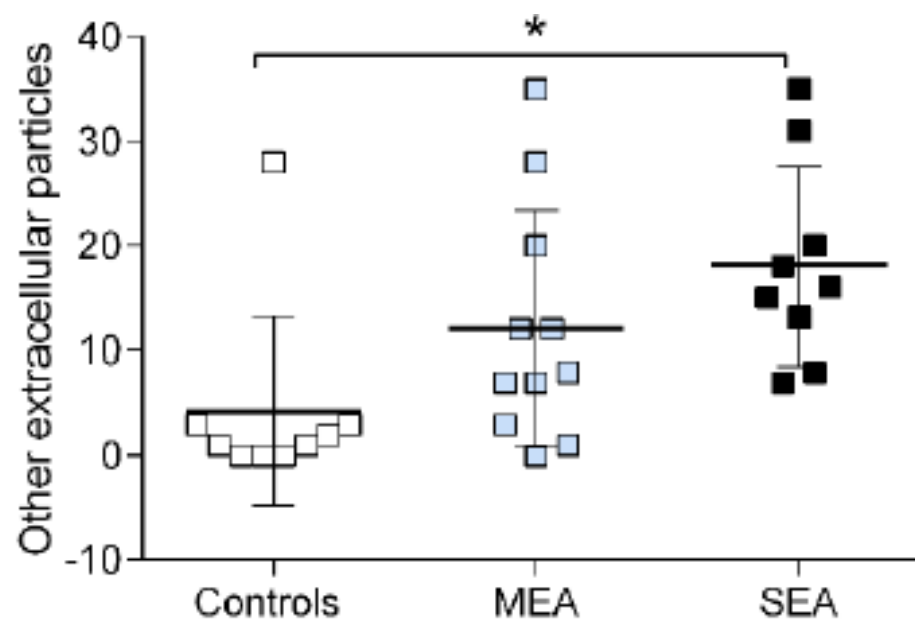
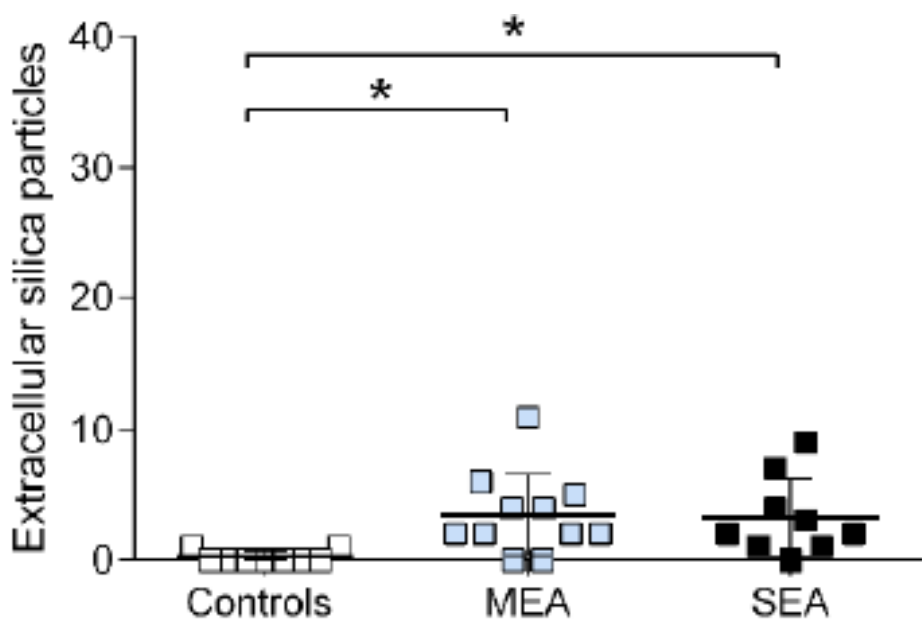


# RESULTS

EXTRACELLULAR 17% OF TOTAL PARTICLES

ALTERED MUCOCILIARY SYSTEM

## SILICA







# RESULTS

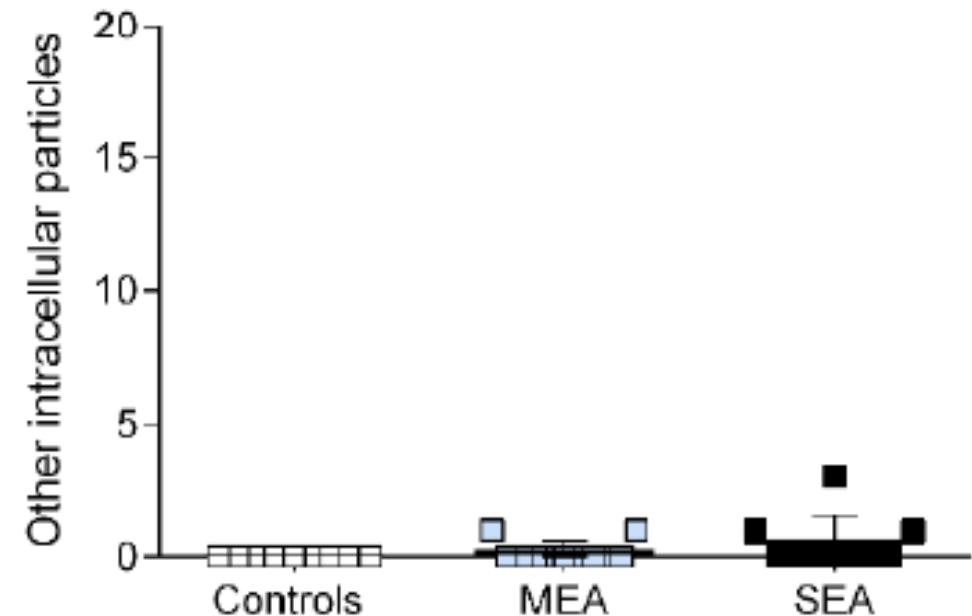
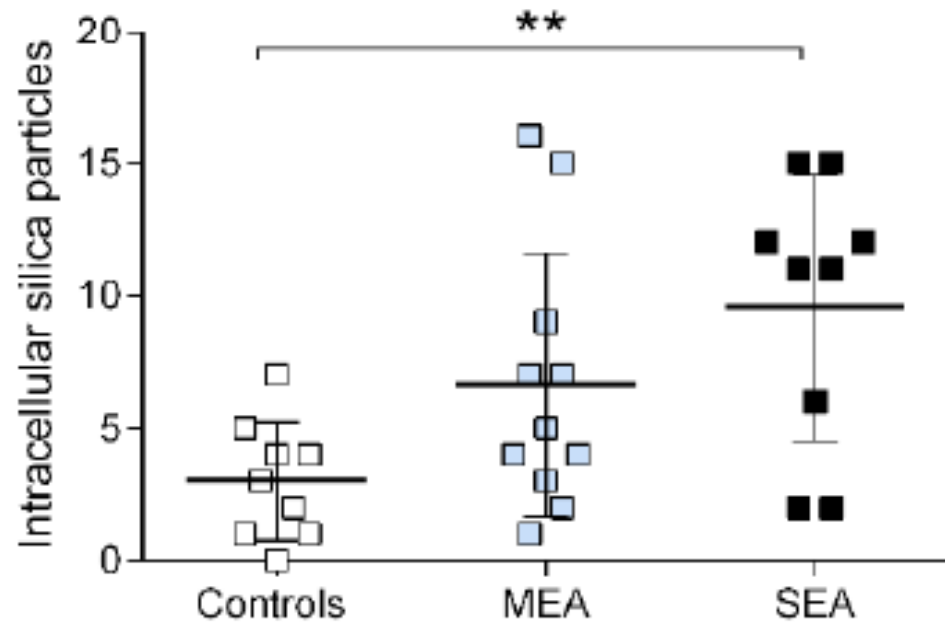
## SILICA

EXTRACELLULAR 17% OF TOTAL PARTICLES

ALTERED MUCOCILIARY SYSTEM

INTRACELLULAR 96% OF TOTAL PARTICLES

**BIOPERSTENCE  
BIODURABILITY  
and  
FIBROSIS**



# RESULTS



**10/42 CASES OBSERVED**

**SILICA PARTICLES → SEA > MEA > CONTROLS**  
**p= 0.05 (Kruskal Wallis Test)**

**SEA**  
**case 1: 20%**  
**case 2: 22%**  
**case 3: 33%**

**MEA**  
**case 4: 0%**  
**case 5: 7%**  
**case 6: 11%**  
**case 7: 14%**

**controls**  
**case 8: 4%**  
**case 9: 9%**  
**case 10: 12%**



# CONCLUSIONS

**INCREASED  
SILICA PARTICLES**

**primary origin**

**secondary origin**

**POSSIBLE  
ROLE  
IN EQUINE  
ASTHMA  
PATHOPHYSIOLOGY**

**ALTERED  
MUCOCILIARY  
CLEARANCE**

**DECREASED  
RESPIRATORY  
FLOW**

**AIR TRAPPING**



# LIMITS

## **It's a Pilot Study!**

**HIGH NUMBER OF VARIABLES AND BIASES**

**WE CANNOT DETERMINATE WITH CERTAINTY  
whether the number of silica particles reaching the alveoli is the cause of  
the onset or progression of asthma or leads exacerbation of clinical signs**

**THE TECHNIQUE USED TO INVESTIGATE SILICA PARTICLES**

# **ACKNOWLEDGEMENTS**

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**GRAZIE PER L'ATTENZIONE!**